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Prioritized Guidelines for Environmental Fate Testing of One Halogenated Hydrocarbon: Chlorobenzene

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Protocols for environmental fate testing of chemicals that are of interest to the Office of Toxic Substances have been divided into five levels of increasing complexity and cost: (1) review and evaluation of available information, (2) development of any missing physical parameters, (3) screening tests, (4) intermediate tests, and (5) detailed laboratory or field tests. Chlorobenzene is used as an example to determine the types of tests to be run. The level of testing required should be based upon the quantity of the chemical being released to the environment, toxicity (if known), and a cost/benefit analysis.

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N O T I C E

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I. INTRODUCTION

In order to assess the potential environmental hazards associated with the commercial use of chemical substances, tests on their environmental fate and biological effects need to be conducted. The number, extensiveness, and cost of these tests should be dependent upon such factors as the quantity of the chemical being used and the likelihood of the chemical being released to the environment. In order to provide a relationship between environmental exposure and the degree of testing, priorities and protocols for the tests are required. This report is a first step in developing such priorities and protocols for determining the fate of chemicals which are the responsibility of EPA's Office of Toxic Substances. In this report, environmental fate is defined as all transport (including bioconcentration) and alteration (including degradation) processes which take place in nature; excluded are any biological effects. The fate of an environmental contaminant has considerable impact upon its potential adverse effects since the environmental fate processes determine the form and quantity of the contaminant that may reach a sensitive biological site.

The case study chemical was selected from the chlorinated hydrocarbon family, a group of chemicals that has provided numerous environmental pollution problems (e.g. DDT, dieldrin and related compounds, PCB's, hexachlorobenzene). Using this example, the types of environmental testing will be discussed and priorities for testing will be suggested. The case study compound, chlorobenzene, provides an example of a high production volume, water insoluble, fairly volatile chemical that is probably released to the environment in significant quantities. However, there are many commercial organic compounds (e.g., gases and solids, and compounds capable of dissociation), as well as

other chemical compounds for which the test case will provide little insight into the type of environmental fate testing that should be undertaken. These compounds need to be considered in future reports.

II. DEVELOPMENT OF ENVIRONMENTAL FATE TESTING PROTOCOLS FOR TOXIC SUBSTANCES

A. Existing Environmental Fate Testing Documents

The Office of Toxic Substances, EPA, has funded two companion studies which have reviewed and evaluated test methods for determining environmental persistence and routes of degradation (Howard et al., 1978) and environmental transport (Mitchenson et al., 1978). These reports provide a good reference to the test methods that are available, their reliability and reproducibility, and relative cost, but little attempt was made to design protocols or prioritize the tests for individual chemicals.

In 1974, Woodard conducted a survey of test methods that were used by industry to evaluate health hazards from individual chemicals. He organized the methods into categories of studies that were conducted in response to four levels of exposure. The levels of exposure with the environmental fate tests that are conducted follows:

Level I Exposure - chemical is still in laboratory or pilot plant production where a limited number of persons may be exposed.

Fate tests: None

Level II Exposure - more persons involved in the production or industrial use of the chemical, but exposure rates are low.

Fate tests: Half-life determination in water and soil, etc.
Biological or chemical oxygen demand
Odor, etc.

Level III Exposure - more individuals persons exposed through occupational, hobby, repair, or incidental chemical contact.

Fate tests: Biodegradation
Metabolic or decomposition products
Transport mechanisms

released to and reside in the environment. However, the hazard involved in unstandardized tests is that considerable amounts of money and effort may be spent in an attempt to determine the environmental fate of a chemical, but result in little insight into the transport and degradation processes if the test is not well designed (for an example of this, see Howard and Saxena, 1978). Also, the interpretation of unstandardized tests frequently varies, which makes it difficult to compare one chemical to another.

B. Chlorobenzene Case Study

Protocols for environmental fate testing of a chemical should reflect an evaluation of all the available information including chemical marketing and physical-chemical data. Chemical marketing data provides insight into the quantities of the chemical that might be released and the medium into which the release will initially occur. This provides guidance for the extent of testing that is justified and the type of tests that should be undertaken. For example, a compound used as a chemical intermediate whose production volume is less than 100,000 pounds probably should only receive inexpensive screening tests, or perhaps no fate testing at all. Depending upon the results from screening tests (e.g., a compound may appear to be very persistent), more testing may be necessary. Compounds used as solvents (probably lost to the atmosphere) should be tested first in the vapor phase, while chemicals that are deposited in landfills should be tested first with soil systems.

Physical and chemical data can be used to estimate stability in the environment and routes of transport (bioaccumulation, evaporation, leaching, etc.). Such estimates will be considered with the test case chemical.

In considering the case study, information on toxicity, environmental monitoring, and environmental fate, although available, has been ignored in order

that the test case be more representative of an existing or new chemical for which the Office of Toxic Substances (OTS) will be called upon to design protocols. Most of the chemical marketing data and chemical and physical properties will be available for all the chemicals reviewed by OTS. Information on toxicity should have an impact upon the degree, but not the type, of environmental fate testing to be conducted. Extremely toxic or carcinogenic, mutagenic, or teratogenic chemicals should be extensively tested for environmental fate. However, rarely will a complete toxicologic evaluation be available and, therefore, the extent and type of environmental fate tests should rely heavily on estimates of the amount released to the environment and physical and chemical properties. Also, toxicologic evaluations would not consider hazardous degradation products which might be formed in the environment.

Ambient monitoring data may be used to establish protocols for existing commercial chemicals, but will be of little use with new products. Monitoring should not be used as a substitute for good environmental fate testing, or vice versa, because frequently chemicals have not been detected until after levels causing adverse effects have been reached and considerable time may be required to eliminate the contamination. Also, intermedia transport (e.g. evaporation from soil) may provide misleading monitoring data concerning the environmental fate of a chemical. Thus, environmental monitoring should be used as a check on environmental fate testing of known chemicals and as a way to identify unrecognized contaminants which may have slipped through the pretesting procedures (e.g. by-products from commercial processes).

The following sections outline a procedure for devising environmental fate testing protocols for chlorobenzene. No attempt has been made to determine

the exact extent of testing that is required, since such details need to be considered relative to other chemicals and in terms of cost-benefit. However, ordering of the tests has been attempted and interrelationships between the tests have been examined.

Step 1 - Review Available Information

Step 1 a - Consideration of Chemical Marketing - Environmental Release Data

Table 1 summarizes the chemical marketing information on chlorobenzene. The chemical is mostly used as a chemical intermediate, which should reduce the amount of the chemical that is released to the environment.

Table 1. Chemical Marketing Data for Chlorobenzene (SRI, 1975)

Chlorobenzene (Millions of Pounds)			
Capacity		659	(1976)
Production		403.5	(1972)
Imports		-	
Exports		-	
Consumption	Phenol Production	110	(1972)
	Chloronitrobenzenes Production	130	(1972)
	DDT Production	20	(1972)
	All Other[a]	144	(1972)
	Total	404	(1972)

[a] Trade literature estimates that at least 50 million pounds per year of chlorobenzene are used as solvents.

However, significant quantities (50 million pounds annually) appear to be used for solvent purposes, which might result in significant release to water systems (if the solvent is used in contact with water) or the atmosphere (during use or distillation recovery). Presumably, more detailed marketing and perhaps environmental release information would be available to the Office of Toxic Substances. However, the available information suggests that environmental fate testing in simulated atmospheric systems and in water should be considered. If some of the solvent applications result in disposal in landfills, soil systems may also be considered.

Tests such as Warburg or model activated sludge systems indicate the treatability of a chemical in an activated sludge water treatment plant. Such tests provide indications of environmental release when a chemical is treated in a biological water treatment plant, but provide only limited information on biodegradability in natural water systems. Thus, such tests should not be considered environmental fate tests.

Step 1 b - Evaluations of Physical and Chemical Properties

Chlorobenzene is a liquid at ambient temperature with a relatively high vapor pressure (9 mm Hg at 20°C), intermediate partition coefficient, and low water solubility (380 ppm). The ultraviolet absorption spectrum in isooctane indicates only very slight absorption above 280 nm (λ_{max} = 271 nm). However, correlations between absorption spectra and photodegradation are poorly understood. Direct absorption of energy is not required for photolysis (e.g. sensitizer reactions). Other physical properties of chlorobenzene are listed in Table 2, but they have little importance to environmental fate. The chemical structure of chlorobenzene (chlorinated

Table 3. Physical Properties of Chlorobenzene (Hardie, 1964)

melting point, °C	-45.21
boiling point, °C	131.5
steam distillation point,[a] °C	90
log partition coefficient[a] (octanol/water)	2.88
water solubility[b]	360 ppm
d_4^t	
0°C	1.1293
10°C	1.1167
16.5°C	1.1058
n_D^t	
15°C	1.52748
20°C	1.52460
surface tension, dyn/cm	
15°C	33.86
20°C	33.28
specific heat, C_p (T = °K)	0.2988 + 0.00074 T
critical temperature, °C	359.2
critical pressure, mm Hg	33962
critical density	0.3654
explosive limits in air, vol %	
lower, 100°C	1.8
upper, 150°C	9.6
flash point, °C	24
dielectric constant	
20°C	5.6493
70°C	6.006
106°C	4.635
at bp	4.144
dipole moment (dil. benzene soln.), esu	1.58×10^{-18}
heat of combustion (at constant pressure), kcal/g-mole	763.88
latent heat of vaporization, kcal/g-mole	8.73
viscosity, cP	
15°C	0.844
30°C	0.711
60°C	0.512
120°C	0.313
130°C	0.292
vapor pressure, mm Hg	
0°C	3
20°C	9
40°C	26
60°C	65
80°C	144
100°C	292
120°C	543

[a] Leo et al., 1971
[b] Metcalf and Lu, 1973

aromatic) suggests that it would be fairly stable in the environment by analogy with other well known contaminants (DDT, PCB's, hexachlorobenzene). Such structure-environmental fate correlations should be used whenever possible (see review in Howard et al., 1975).

The physical properties can be used to estimate the rates of transport of a chemical that is released to the environment. These calculations for chlorobenzene are summarized in Table 3, where calculated values for benzene and DDT are provided for comparison. The calculations indicate that chlorobenzene has a low potential for bioaccumulation and should evaporate relatively fast from water (C_{H_2O}/C_{air} = 100 is defined as a chemical that is rapidly lost from water surfaces). The calculated values should be used with caution since they are only estimates that may give misleading impressions. For example, although the calculated ecological magnification for chlorobenzene is 9, the experimental value is 645 (Metcalf and Lu, 1973), which is comparable to the experimental value for tetrachlorodibenzodioxin (calc. = 25,000; exper. = 822) and hexachlorobenzene (calc. = 20,000; exper. = 343).

Nevertheless, the physical properties can provide considerable insight into the type of testing that should be considered. With chlorobenzene in water, it appears that a major loss mechanism may be volatilization. Whether degradation processes in water can compete will have to be determined experimentally. The calculations also emphasize the importance that atmospheric processes are likely to play in the environmental fate of chlorobenzene.

Step 1 c - Review Literature on Chemical Reactivities and Product Formation

Literature searches are considerably cheaper than experimental work, and, therefore, the former should be undertaken before any

Table 3. Calculations of Bioconcentration and Evaporation Rates from Physical Properties

	Bioconcentration Factor Calculated From Partition Coefficient[a]	Ecological Magnification Calculated From Water Solubility[b]	Half-life (Hours) of Evaporation From Water Calculated From Vapor Pressure, Water Solubility, and Molecular Weight[c]	Ratio C_{aq}/C_{air} Calculated From Vapor Pressure and Water Solubility[d]
Chlorobenzene	46	9	5.6	0.22
DDT (for comparison)	430	29,300	73.9	326
Benzene (for comparison)	29	4	4.81	4.5

[a] Neely et al. (1974)
[b] Metcalf and Lu (1973)
[c] Mackay and Leinonen (1975)
[d] Appendix VI Guidelines for Registering Pesticides

experimental work begins. Information may already be available that would eliminate the necessity for some or all (highly unlikely) of the environmental fate tests. Also, examination of previous studies will provide insight into analytical methods that might be used and breakdown products that might be formed.

A preliminary examination of available information on chlorobenzene indicates that its degradation in aerated lagoons (Garrison, 1969) and activated sludge has been examined and its photolysis (wavelength less than 253 nm - not representative of sunlight) in the gas phase studied (Ichimura et al., 1973). Also, Gibson et al. (1968) found that chlorobenzene is converted to 3-chlorocatechol when it was incubated with Pseudomonas putida grown with toluene as a sole source of carbon; this information will assist in the identification of degradation products. In addition, as previously mentioned, the ecological magnification in a model ecosystem has been determined (Metcalf and Lu, 1973).

Step 2 - Development of Any Missing Physical Parameters

After reviewing the available information, there may be some missing physical parameters (e.g., water solubility, partition coefficient, vapor pressure, dissociation constant). The partition coefficient may be obtained from the tabulation of Leo et al. (1971), calculated using additivity principals (Hansch et al., 1972), or measured experimentally (see Pesticide Registration Guidelines Appendix VI for methodology). The other physical properties will have to be measured experimentally (see Appendix VI). With chlorobenzene, a dissociation constant is unnecessary since chlorobenzene does not dissociate. The water solubility, vapor pressure, and partition coefficient are available.

These physical properties should be measured before experiments on degradation and transport are begun so they can be used in designing the experimental test conditions.

Step 3 - Screening Tests

Tests can be divided into three categories by analytical method: (1) tests that require no analytical method for the chemical (e.g. BOD, Warburg - both measure consumed oxygen) (this lack of analytical method only applies to degradation tests, not transport studies) (2) tests that require specific analytical methods for the chemical, and (3) tests that use radiolabelled material. The use of radiolabelled chemicals for environmental fate testing is the most desirable because it allows for a mass balance of the test chemical and products, is very sensitive, and combined with thin layer chromatography can provide insight into the breakdown products. Chemical analytical methods allow the investigator to follow the disappearance of the parent compound and, if he knows what products to expect, to develop analytical methods for the degradation products. In some instances, radiolabelled parent compound may not be available or an analytical method needs to be developed for the degradation products, and the cost of synthesis or analytical method development are not justified. In these instances, a variety of screening tests may be considered. Examples of such tests are:

BOD
Warburg
River die-away
7-day hydrolysis and photolysis studies under acidic, neutral, and basic conditions (see Wolfe et al., 1976)
Soil adsorption or TLC studies
Rapid evaporation studies from water

Rapid screening tests such as these usually measure the relative increase in oxygen uptake or the disappearance or transport of the test chemical. Long-path infrared cells, which have been used to study atmospheric photooxidation, might also be considered in this category since an analytical method does not need to be developed (infrared absorption). However, the equipment is so expensive and complicated that it should not be considered a screening test.

Screening tests have the advantage that they are rapid and inexpensive. However, they provide very limited information on the environmental fate of a chemical. With BOD and Warburg, the parameter being measured, oxygen uptake, is only an indirect indication of degradation. The results are difficult to interpret unless extremes, such as no oxygen uptake or 100% of the theoretical oxygen demand are found, which is rarely the case, and the conditions of the Warburg test are more similar to an activated sewage treatment plant than to natural conditions. The die-away and hydrolysis tests only determine the rate of disappearance of the test compound (good indication of persistence), but provide little insight into the degradation products, which is equally important.

With chlorobenzene, if screening tests are the only ones justifiable, a BOD and river die-away test probably should be run, and perhaps a rapid photolysis study in hexane (provide preliminary indications of the atmospheric photooxidation). Since chlorobenzene is very stable in water, hydrolysis studies do not seem justified. The Warburg test indicates treatability rather than environmental fate and, therefore, should not be undertaken unless it is anticipated that sizable quantities of chlorobenzene will pass through an activated sludge treatment plant. Because of the limited information provided from the screening tests, it seems reasonable to consider avoiding these tests if more detailed studies are anticipated. Also, 100 uCi of C^{14} uniformly labelled

chlorobenzene is available for about $80, so it would seem more reasonable to conduct any screening tests with radiolabelled material (BOD and Warburg are exceptions).

Step 4 - Intermediate Tests

Tests in this category, with the exception of atmospheric photo-oxidation systems, are generally characterised by the use of radiolabelled material. The difference between this category and the following category of more detailed studies is that any major (> 10% - Appendix VI Pesticide Guidelines) degradation products may be characterised (e.g., TLC mobility, gas chromatography retention time, UV absorbance on TLC (+ or -), radioactive content, or possible TLC spray reactions - Pesticide Guidelines Appendix VI), but not chemically identified. Rigorous and complete chemical identification is extremely time consuming and expensive, although the use of GC-MS systems has considerably simplified the process. Environmental fate tests that fall into an intermediate category include the following:

Atmospheric
- Long-path infrared cells
- Plastic containers
- Glass flask reactors

Water
- Hydrolysis studies (various pH and temperature - determine the kinetics)
- Photolysis with simulated sunlight in various matrices (determine kinetics - use sensitisers and natural water)
- Die-away tests with radiolabelled material (sea, lake, river water from various sources to provide a wide spectrum of microbes, pH, pollution, organic content, etc. - include with and without sediment, sterile control, aerobic and anaerobic conditions)
- Shake culture tests
- Shake culture using pure cultures
- Model sewage treatment systems

Soil

Soils incubated with test chemical (aerobic, flooded, anaerobic conditions, soils suspended in water)
Pure cultures isolated from soil
Greenhouse studies

Transport

Volatility - from water (Dilling et al., 1975)
Bioaccumulation and biomagnification - terrestrial-aquatic or aquatic model ecosystems (Metcalf method, Isensee method)
Uptake by individual biota
Desorption from organisms

The above list illustrates the considerable number of laboratory tests that may be undertaken to determine the environmental fate of a chemical. Not included are the many variations of the above tests that are possible (see Howard et al., 1975; Witherspoon et al., 1976). Some techniques, such as the use of pure culture, might more appropriately be used to determine the pathways of degradation (next category). Results using pure culture, although they have been used to indicate persistence (ability to use a chemical as a carbon and energy source), are difficult to relate to the rates of degradation that might take place in nature.

Without considering a particular compound, it is extremely difficult to prioritize the above tests. With chlorobenzene, it would appear to be important to determine the rate and products of atmospheric photooxidation, the rate of evaporation from water, and the rate and products of degradation (chemical and biological) in water. If the rate of degradation in water is slow compared to the rate of evaporation, studies of the atmospheric photooxidation will be considerably increased in importance. Products of atmospheric photooxidation (e.g. perhaps chlorophenols) are probably less volatile, suggesting that they should be studied in water or soil systems, although considerable insight into the behavior of chlorophenols in soil and water is already available. Long-path infrared cells for atmospheric tests have the advantage that no analytical method

needs to be developed. However, the equipment used for atmospheric studies is not available in many laboratories, so plastic or glass containers may be preferred. The analytical method used, the scheduling of tests, etc. should be left to individual researchers.

Step 5 - Detailed Laboratory and Field Tests

Studies in this category include techniques oriented at chemical identification of intermediates, determination of pathways of degradation, tests that examine in detail the effect of various conditions on reaction rates (e.g. the effect of NO_x concentration on photooxidation rates - usually conducted in smog chambers), and field tests. In general, studies in this category are very expensive and time consuming. Detailed laboratory or field tests include:

Air
- Smog chamber
- Release and monitoring studies

Water
- Die-away tests in treated ponds, lakes, streams
- Treatability of a chemical in a commercial activated sludge treatment plant
- Pure culture techniques to determine pathways of degradation

Soil
- Field plots - monitoring
- Landfills - monitoring gases and leachate
- Pure culture techniques to determine pathways of degradation

Transport
- Field studies followed by monitoring biota, soil, water, etc.

Field studies suffer from low reproducibility because of the variable test conditions, but provide the only results where natural conditions are used. However, because radiolabelled material cannot be used, rarely are degradation products examined, although they may be analysed if the field test follows the intermediate test, since the intermediate test may provide indications of the degradation products that will be formed. It seems unlikely that techniques in this

category should be used except for chemicals that are released to the environment in very large quantities (e.g. pesticides - see Appendix VI, detergents, etc.). If justified from release estimates, chloroquenesene should be tested in a smog chamber and perhaps in a die-away test in a pond or lake. Stable metabolites, which may have been determined from the intermediate tests, should also be monitored.

III. CONCLUSIONS AND RECOMMENDATIONS

Establishing protocols for environmental fate testing of chemicals that are of interest to the Office of Toxic Substances has been divided into five steps:

Step 1 Review and evaluation of available information

Step 2 Development of any missing physical parameters

Step 3 Screening tests

Step 4 Intermediate tests

Step 5 Detailed laboratory or field tests

This is only a preliminary result which may need considerable revision before implementation of test protocols. The tests recommended for one test case chemical, chlorobenzene, are considered based upon commercial considerations and chemical and physical properties. More experience is needed with a greater variety of chemical compounds. The level of testing required should be based upon the quantity of the chemical being released to the environment, toxicity (if known), and a cost/benefit analysis.

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